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**IN THE UNITED STATES PATENT AND TRADEMARK OFFICE**

Application of: Eva BINGGELI, et al.  
Serial No.: 10/505251  
Filed: 19.Aug.2004  
Examiner: Helen PRATT  
Art Group: 1794  
Title: **BRASSICA SEEDS**

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Per Telefax: 571 273-8300

Commissioner for Patents  
P.O. Box 1450  
Alexandria, VA 22313 - 1450

Dear Sir;

**AFFIDAVIT UNDER 35 U.S.C. 1.132**

I, the undersigned Eva BINGGELI, a co-inventor of the indicated application, do attest as follows:

- 1) I hold both a Master's degree in Chemical Engineering (Sweden) and a Master's degree in Biotechnology (Switzerland).
- 2) Prior to my present position, I had been employed as a researcher at Pharmacia (Sweden), Enzymlabor Kantonsspital St.Gallen, (Switzerland), and at Sandoz AG, Basel(Switzerland)

- 3) For the past 30 years I have been employed at the research laboratories of Givaudan Flavors Corporation, and my current title is Research Investigator
- 4) I am a co-inventor of an invention "BRASSICA SEEDS" filed at the United States Patent Office under Serial No. 10/505251.
- 4) Under my supervision, the following testing was performed:

Example 1 - Roasting Whole Brassica Seeds

40 g of whole *Brassica juncea* seeds were roasted in a Probat Sample Roaster (PRE 1Z, Probat-Werke von Gimborn Maschinenfabrik GmbH, Germany) at 135- 150°C for 6 minutes. The resulting seeds had an aroma of pleasant, roasted mustard, as determined by a panel of 3 trained people.

Example 2 - Roasting Crushed Brassica Seeds

40 g of crushed *Brassica juncea* seeds were roasted in a Probat Sample Roaster (PRE 1Z, Probat-Werke von Gimborn Maschinenfabrik GmbH, Germany) at 125°C for 3 minutes. The resulting seeds had a bitter pungent mustard like aroma, as determined by a panel of 3 trained people. This aroma was due to the presence of thiocyanates and isothioates.

It was not possible to heat the crushed Brassica seeds above 125°C for 3 mins because the crushed Brassica seed stuck to the roaster wall and burnt. This was because the oil released from the seeds on crushing quickly heated and burnt the surface of the crushed pieces.

The above experiments were carried out on a small scale for health and safety reasons. Roasting large quantities of crushed Brassica seeds is not permitted for health and safety reasons due to the release of thiocyanates and isothiocyanates which are irritants.

### Conclusions

The aroma of the product produced by roasting whole Brassica seeds was determined as pleasant roasted mustard. This is surprisingly different from the aroma of the product produced by roasting crushed Brassica seeds which was determined as bitter pungent mustard like.

This is because on crushing the Brassica seeds the sulphur containing glucosides and glucosinolates, found in the Brassica seeds, were exposed to an enzyme (found predominately just beneath the surface of the seed) which broke them down resulting in the formation of strongly tasting substances, predominately thiocyanates and isothiocyanates, which impart a bitter pungent taste.

This did not occur in the brassica seeds that were roasted whole because the enzyme responsible for the formation of the strongly tasting compounds was denatured before the seeds were crushed.

Further to the above, it was also found that it was not possible to roast crushed Brassica seeds at a temperature of 125°C for more than 3 minutes. On this basis it can be assumed that it is not possible to roast crushed Brassica seeds at a temperature of 160°C for at least 10 minutes without burning them.

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like

so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Respectfully;

Eva Binggeli  
Eva BINGGELI

Date: April 27, 2010

c/o

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CERTIFICATION OF TELEFAX TRANSMISSION:

I hereby certify that this paper and all attachments thereto is being telefax transmitted to the US Patent and Trademark Office to telefax number: 571 273-8300 on the date shown below:

Andrew N. Parfomak  
ANDREW N. PARFOMAK

27. April, 2010  
Date: